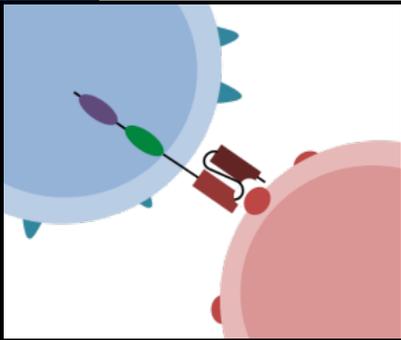


# CAR-T Cells with Enhanced Metabolic Fitness due to Co-expression of Mutant PGC-1 $\alpha$

*Selectively targeting PGC-1 $\alpha$  post-translational modifications enhances or prevents degradation of metabolic fitness in CAR-T cells. Targeted mutations include: Akt phosphorylation of PGC-1 $\alpha$  on serine suppresses PGC-1 $\alpha$  activity; GSK3 $\beta$  ubiquitinates PGC-1 $\alpha$  for proteasomal degradation; Clk-2 phosphorylation of PGC-1 $\alpha$  on multiple serines suppresses PGC-1 $\alpha$  activity; and S6K phosphorylation of PGC-1 $\alpha$  on serine suppresses PGC-1 $\alpha$  activity. PGC-1 $\alpha$  mutant expression during transduction and expansion produces more central and effector memory CAR T cells and reduces short lived effector cells. PGC-1 $\alpha$  mutant expression during CAR T activation increases expression of transcription factors associated with metabolic fitness and maintenance of memory phenotypes.*



## COMMERCIAL OPPORTUNITY

- Several factors are associated with CAR T efficacy. Proliferative capacity and asymmetric division of memory and naïve like phenotypes is required to supply enough cells to eradicate tumor cells and mediate disease remission. Greater persistence results in a reservoir of tumor specific T cells to surveil for disease over time increasing durability of response. Greater metabolic fitness allows CAR-T cells to perform cytolytic and secretory function in a nutrient depleted microenvironment under high oxidative stress. The cells produce ATP via oxidative metabolism to slow or prevent differentiation toward terminally differentiated or exhausted effector phenotypes. The cells also have reduced glycolytic flux. Persistent memory cells are characterized as having increased mitochondrial biomass with tubular morphology and a greater use of oxidative metabolism that relies more on the TCA cycle and ETC to produce ATP.
- PGC-1 $\alpha$  is a 798 a.a. transcriptional coactivator with no DNA binding domain or enzymatic activity. As a coactivator, it can bind with a broad set of transcription factors and induce the upregulation of many complex transcriptional programs. These transcriptional programs combine to initiate mitochondrial biogenesis, enhance oxidative metabolism (Fatty Acid Oxidation, TCA cycle, and Electron Transport Chain (ETC)), enhanced mitochondrial flux (mitophagy), and reduce oxidative stress. A multitude of post translational modifications (PTM) determine PGC-1 $\alpha$  localization, activity, and rate of proteasomal degradation.
- The marketplace is attractive for CAR-T development, as Novartis received approval in August 2017 for Kymriah, its anti-CD19 CAR-T therapy for pediatric B-cell ALL. The trial had an overall response rate of 82.5% (52/63). Although the list price for Kymriah is \$475,000 for a one-time treatment, Novartis has said only those patients who respond by the end of the first month will need to pay. In October 2017, Gilead's Yescarta, an anti-CD19 CAR-T, was approved for large B-cell lymphoma and is listed at \$375,000. In 2017, Gilead acquired Kite Pharma for \$11.7B, and in 2018, Celgene acquired Juno Therapeutics for \$9B. Juno is also developing a CD-19 CAR-T therapy.

## TECHNOLOGY

Co-transduction of CD8 T cells with CAR encoded retrovirus and mutant PGC-1 $\alpha$  encoded retrovirus increases the percentage of central memory CAR cells (Tcm) and effector memory CAR T cells (Tem) present after transduction. Co-transduction of CD8 T cells with CAR encoded retrovirus and mutant PGC-1 $\alpha$  encoded retrovirus decreases the percentage of effector CAR T cells (Teff) present after transduction. Tcm and Tem phenotypes were quantified via flow cytometry after 7-10 day transduction and isolation of CAR + and mutant PGC-1 $\alpha$  + T cells using flow cytometry based cell sorting. The PGC-1 $\alpha$  mutants were resistant to deactivating phosphorylation by Akt and resistant to ubiquitination by GSK3 $\beta$  and subsequent proteasomal degradation.

## PUBLICATION/PATENT

- Provisional Patent filed on November 25, 2019 for Drs. Locke and Atkins.

### CONTACT

Haskell Adler PhD MBA  
Senior Licensing Manager  
Haskell.Adler@Moffitt.org  
(813) 745-6596

### LICENSING OPPORTUNITY



19MB066.2020.01